Pre Ph. D. Registration Synopsis

Comparative Morphohistochemical and Antimicrobial Studies on some Eusporangiate Pteridophytes of Ranchi District of Jharkhand

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State of Art:

Pteridophytes by the virtue of their possessing great variety and fascinating foliage have drawn the attention and admiration of horticulturists and plant lovers for centuries.[1] Pteridophytes have complex leaves with elaborate shapes.[2] They are represented by about 305 genera, comprising more than 10,000 species all over the world. [3] About 191 genera and more than 1,000 species are reported from India.[4]

In Pteridophytes sporophyte is the dominant generation. Unlike the seed producing plants of angiosperms and gymnosperms, pteridophytes reproduce with the help of spores produced in sporangia. Among them eusporangiate represent a highly diverse subclass characterized by the type of sporangia arise from several epidermal cells and not from a single cell as in Leptosporangiate Pteridophytes. Eusporangiate Pteridophytes bear relatively massive sporangia that contain thousands of spores that may release passively or actively.[5] Alternation of gametophyte and sporophyte can be understood from morphological perspective.[6]

These vascular cryptogams are plants that evolved from aquatic to terrestrial ecosystem. Pteridophytes thrive in a variety of ecological habitat, particularly in shaded environment making them a significant component of forest ecosystem. The study area Ranchi district of Jharkhand is known for its rich Pteridophytic diversity attributes to its unique geographical and tropical climatic conditions, which favors the growth of this pteridophytic vegetation. Total 33 Pteridophytes are reported from the study area.[7] Apart from their taxonomic and ecological significance Pteridophytes remain underexplored in their terms of Morphohistochemical characteristic and medicinal applications.[8] Histochemistry is an essential analytical tool use to decipher specific physiological and study plant cell structure.[9] Many of the Pteridophytes have medicinal properties and frequently used by the tribal people for curing different types of disease.[10] They possess many useful phytochemical and may be by the presence of these phytochemicals these Pteridophytes shows antimicrobial and antifungal properties.[11] Human beings have been using Pteridophytes because of their several useful properties, including food and medicine.[12] Several species have been traditionally used in indigenous medicine for treating infections, wounds and inflammatory conditions.[13] However there is limited scientific validation of their antimicrobial efficacy, especially for eusporangiate Pteridophytes of Ranchi district.

This study aims to conduct a comparative morphohistochemical and antimicrobial studies of selected eusporangiate Pteridophytes of Ranchi district. Anatomical studies provide essential insights into their structural adaptations, taxonomy and evolutionary traits, which can be useful for species identification and understanding their ecological strategies. The histochemical detection is based on the simple principle that when a tissue or cell is exposed to a staining solution it will react specifically by forming a coloured insoluble and product detectable by microscopy.[14] Contrast in plant tissues, with specific colouring reagents allows metabolites characterization such as flavonoids, alkaloids and others.[15] By correlating anatomical features with bioactive potential, this research will contribute to both plant taxonomy and pharmacological advancement. The findings will enhance our understanding

of Pteridophyte biology and highlight their potential application in the field of taxonomy, morphohistology and pharmacognosy.

Problem to be investigated:

- Morphological Pteridophyte study – Morphological investigation along with spore morphology to identify the Pteridophyte and to understand how closely species are related.
- Histochemical study – Detailed Anatomical study that involves the use of specific staining reactions and chemical assays to identify and localize various biochemical compounds and cellular structures within plant tissues of pteridophyte.
- Extraction and characterization of secondary metabolites.
- Selection of relevant bacterial and fungal strains.

Objectives:

1. To identify and classify selected eusporangiate Pteridophyte *Equisetum arvense, Ophioglossum reticulatum, Selaginella bryopteris* collected from different areas of Ranchi district.
2. To study the morphohistochemical characteristics including leaf, stem, and rhizome anatomy stained with different reagents.
3. To perform phytochemical analysis to detect the presence of bioactive compounds.
4. To screen the antimicrobial activity of pteridophyte extracts against selected bacterial and fungal strains.

Motivation:

Pteridophytes, particularly the eusporangiate represent an ecological and medicinally significant group of plants, yet their morphohistological adaptations and antimicrobial properties remain largely unexplored.[16] The Ranchi district of Jharkhand, favors the growth of pteridophytic vegetation which are traditionally used by the tribal population of the study area from the ancient times. However their documentation and scientific validation of their medicinal potential, particularly their antimicrobial efficacy is still limited.[17] Pteridophytes with their unique secondary metabolites offer promising alternative for developing new antimicrobial agents.[18] Additionally, understanding the morphological and anatomical features of these Pteridophytes at the histological level can provide discernment of their identification, classification and medicinal properties.

Hypothesis:

Proposed investigation will help better to understand the specific structural and anatomical features of these eusporangiate which are associated with the production of bioactive compounds effective against certain pathogens. Testing this hypothesis would involve detailed morphological and histochemical studies along with antimicrobial efficacy.

Research Design & Methodology:

The study involves a comparative morphohistochemical and antimicrobial analysis of selected eusporangiate Pteridophytes of Ranchi district. The methodology includes collection, identification, anatomical study, phytochemical analysis and antimicrobial screening to establish correlation between structural features and bioactive potential.

1. ***Collection and Identification of Pteridophytes***
    - Pteridophytic specimens will be collected from different habitats including forests, wetlands and shaded rocky surface.
    - Identification will be performed using standard floras and taxonomic keys (e.g. H. H. Hains, Beddome and Pteridophytes of India)
    - Voucher specimen will be prepared, preserved and deposited in a recognized herbarium for further reference.

2. ***Morphohistochemical Studies***
    - Macroscopic Study–Morphological features such as frond type, rhizomes, Venation pattern and spore characteristics will be examined.
    - Microscopic Study –
        - Section Cutting–Section of leaf, petiole and rhizome will be prepared using a microtome and study of different types of tissues.
        - Staining–Specific stains are used to react with the target chemical compounds, creating coloured product that can be visualized under a microscope.
        - Maceration–will be used to analyse fibers, vessels, traceids etc.
        - SEM–will be used to study the spore morphology.

3. ***Preparation of Plant Extracts:***

    Preparation of plants extract will be done through the cold and hot maceration methods on the basis of availability.

4. ***Phytochemical Analysis:***
    - Preliminary screening for bioactive compounds such as alkaloids, flavonoids, tannins, saponins and Phenolies will be done using the standard methods.[19]
    - Quantitative estimation of total bioactive compounds using UV – Visible Spectrophotometry.[20]

5. ***Antimicrobial Screening:***
    - Antimicrobial Screening by using the test organisms like *E. coli*, *Staphylococcus aureus*, *Candida albicans* and *Aspergillus niger* etc.
    - It involves testing the effectiveness of compounds of plant extract using standard protocol of either Disc diffusion method or Well diffusion method.[21]

Tools to be used:

1. For collection of plant specimens
    - Pruning Shears
    - Trowel or Shovel
    - Plastic Bags
    - Field Notebooks
    - Plant Press
    - Tags
    - Camera

2. For Morphohistochemical study
    - Light Microscope
    - Microtome
    - SEM
    - Glassware
    - Different staining reagents

3. For Phytochemical Screening
    - Chemical Reagent (Mayer's reagent, Ferric chloride etc)
    - TLC (Thin Layer Chromatography)
    - UV – Visible Spectrophotometer

4. For Antimicrobial Analysis
    - Laminar Air flow chamber
    - Autoclave
    - Petri plates and culture tubes
    - Incubator
    - Sterile Pipettes and Micropipettes
    - Microbial strains

Significance of the Study

The study of eusporangiate provides significant importance in the field of Taxonomy, anatomy and ecology of Pteridophytes. Investigating the morphohistochemical characteristics of these Pteridophytes enhance our understanding of plant diversity and evolutionary relationship. By screening for antimicrobial properties, this research may identify new bioactive compounds which are effective against the various Pathogens, contributing to the development of new drugs.

Its relevance to the present day problems and need of the society

The present study is highly relevant in the context of Modern taxonomy, classification and evolution of Pteridophytes. Pteridophytes contain many bioactive secondary metabolites that having antimicrobial and antifungal properties. Proper documentation and scientific validation of these medicinal Pteridophytes can help integrate Pteridophyte based medicine into modern pharmacology.

Contribution to the existing knowledge

This investigation adds valuable information to the botanical literature in the field of Taxonomy, Anatomy and Pharmacognosy of Pteridophyte. The study provides a deeper understanding of specific structural characteristics that may influence to the production of new bioactive compounds that may be used in the formulation of new drugs.

Future work

Detail investigation of Pteridophytes flora of Ranchi district at molecular level.

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